

SPECIES DISTINCTION IN ABDOMINAL PIGMENTATION PATTERNS BETWEEN FEMALES OF *DROSOPHILA MELANOGASTER* AND *D. SIMULANS*, FROM A SPANISH POPULATION¹

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ABSTRACT: The sibling species *Drosophila melanogaster* and *D. simulans* coexist in natural conditions. Whereas males are easily recognizable by their genital arches, females were considered to be indistinguishable but for their eye sizes. In many papers separate female counts were omitted because of this difficult characteristic. However, the abdominal pigmentation pattern was found to be different between the two species in a Spanish population. The discrimination of the females based on pigmentation differences was checked by electrophoresis and found to be very reliable.

Since the discovery of Sturtevant (1919) that *Drosophila melanogaster* has a closely resembling sibling species *D. simulans*, both species are known to be cosmopolitan and coexistent (Lachaise et al., 1988). In some population screens the authors make no effort to distinguish the females of the two species, and only mention their grand total (Tantawy & Soliman, 1967; references in Lachaise et al., 1988). Most often, research starts with isofemale lines and checking their progeny in which the males of the two species are distinguishable due to different genital arches (Burla, 1951; Coyne, 1983; Sturtevant, 1919). Based on measurements of eye sizes of *D. melanogaster* and *D. simulans*, it is possible to make a distinction between the females (Burla, 1951; Gallo, 1973; McNamee & Dytham, 1993) but it is a painstaking job when large numbers of flies have to be examined. Okada (1956) described a way of discrimination based on differences in egg guides, but this character also necessitates much practice to distinguish the two species. A high number (up to 45 %) of misqualifications of *D. melanogaster* have been reported, based on different eye size definitions (McNamee & Dytham, 1993 and references therein).

We used flies captured in traps in Carboneras (Almería, Spain) to see if a way of morphological distinction by abdominal pigmentation differences (Gallo, 1973) might be applicable in our population of *D. melanogaster* and *D. simulans*. Eye size was used as the character to separate the species, but we also checked the pigmentation of the sixth tergite to see whether a useful correlation existed.

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Females emerging from *Opuntia ficus-indica* fruits were primarily discriminated by the pigmentation pattern of the sixth tergite. All flies deemed *D. melanogaster* were subjected to electrophoresis for other reasons (Eisses and Santos, 1997).

MATERIALS AND METHODS

Flies were captured with mashed banana traps during five days in the Carboneras area (Almería, Spain; 37°00'N; 1°53'W) and locations nearby (Eisses and Santos, 1997). *Opuntia ficus-indica* fruits (prickly pears) were put in trays in a semi-abandoned *O. ficus-indica* plantation and left for almost seven days in the field. After recollection, the fruits were placed in glass jars, and emerging flies were aspirated. Captured and emerged flies were checked for *D. melanogaster* morphology and frozen at -29 °C until electrophoresis.

ADH is a diagnostic enzyme between *D. melanogaster* and *D. simulans* because of clearly distinctive bands in gel electrophoresis (Eisses, Van Dijk & Van Delden, 1979).

RESULTS

The apparent *D. melanogaster* females trapped in the *O. ficus-indica* plantation near Carboneras were separated from *D. simulans* by eye size only, whereas flies from the other locations were separated at the species and sex level by eye size and genital arches. After electrophoresis the number of misqualifications of *D. melanogaster* was calculated (Table I A). Almost 21 % of the female flies turned out to be *D. simulans* (Table I A 1). For females and males together a general misqualification of 11 % was obtained (Table I A 2).

Table I. Number of flies initially separated as *D. melanogaster* and percentage of misqualifications based on electrophoresis of flies trapped in banana baits in a semi abandoned *O. ficus-indica* plantation (A 1) and other locations close to Carboneras (A 2), and of flies emerging from *O. ficus-indica* fruits collected at the plantation (B).

Method of distinction	No. initially separated as <i>D. melanogaster</i>	% actually determined as <i>D. simulans</i>
A 1 Eye size	226 females	20.8
2 Eye size / Genital arch	437 females / males	11.0
B Pigmentation of 6th tergite and eye size	1078 females	1.68 ± 0.26*
Genital arch	1092 males	0.64 ± 0.034

*Empirical Standard Deviation

Flies emerging from *O. ficus-indica* fruits were separated primarily by the morphological distinction of the pigmentation pattern of the sixth tergite (Fig. 1) and in cases of doubt the eye size was examined as well. Approximately equal numbers of female and male flies were checked by each of us. After checking the flies with electrophoresis, the average percentage of misqualifications of the females was calculated to be $1.68\% \pm 0.26$. This is in the same order as misqualifying male flies (Table I B).

The most important difference between *D. melanogaster* and *D. simulans* females is the black pigmentation of the sixth tergite, which runs to the ventral margin in *D. melanogaster*, whereas the pigmentation border line in *D. simulans* makes an angle with the tergite margin. It forms a continuous line with the pigmentation border line in the seventh tergite (Fig. 1 a). In contrast with an apparently monomorphic *D. simulans*, we observed large variation in abdominal pigmentation patterns in this natural population of *D. melanogaster* and also in some laboratory strains (Fig. 1 b - i).

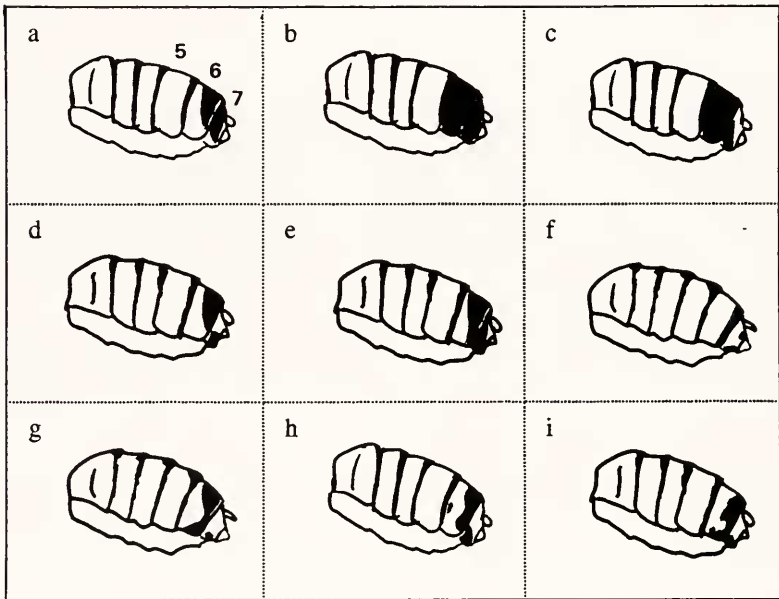


Figure 1. Pigmentation patterns of the 6th and 7th tergite of *D. simulans* (a) and *D. melanogaster* (b - i). Within *D. melanogaster* variation was present in wild type populations and in homozygous or isogenic laboratory strains Groningen SSN (b) and Groningen FFF (d). None of the *D. melanogaster* strains was monomorphic.

We have demonstrated that distinction between females of a natural population of *D. melanogaster* and *D. simulans* in Spain can be made in an easy and reliable way. As similar observations have been made on females from a Brazilian population (Gallo, 1973) and from a midwestern U.S. population (Thompson, Hisey & Woodruff, 1979) it might be generalized to more populations of *D. simulans*. It seems worthwhile to excavate information about other *D. simulans* populations with respect to female abdominal pigmentation of the sixth tergite to establish whether or not *D. simulans* is world wide monomorphic for this character in contrast to *D. melanogaster* (Robertson, Briscoe & Louw, 1977; David, Capy & Gauthier, 1990). Robertson, Briscoe and Louw (1977) described the focus *fap* (female abdomen pattern) to be residing on the extreme tip of the 3L chromosome, with some effects from the fourth chromosome. This might be the reason why the *D. melanogaster* Groningen-FFF strain, used as a reference in electrophoresis, showed a pigmentation pattern in the sixth tergite like *D. simulans* (Fig. 1d). This strain is partly homozygous for the second chromosome and the third chromosome.

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CURTIS W. SABROSKY

The American Entomological Society deeply regrets the recent passing of Dr. Curtis W. Sabrosky, a friend and Honorary Member of the Society. Although a long time member of the Entomological Society of Washington, DC, in recent years, following his move to Medford Leas, Medford, NJ, Curtis regularly attended meetings of the American Entomological Society.

Because Curtis was honored in 1982 in a "festschrift" (Vol. 10) edition of the *Memoirs of the Entomological Society of Washington*, only a brief notice is now planned by the Washington Society, together with publication of a complete bibliography of his entomological contributions.

— H.P.B.